

Patent claims

1. An in vitro method for the determination of the formation of endothelins in serious diseases, in particular cardiovascular diseases, inflammations, sepsis and cancer, in whole blood, plasma or serum of a human patient for purposes of medical diagnostics, wherein the formation of endothelin-1 (SEQ ID NO:2) and big endothelin-1 (SEQ ID NO:3) is determined by determining those C-terminal fragments of preproendothelin-1 (SEQ ID NO:1) which are recognized by antibodies which bind to peptides which correspond to peptide sequences in the range of amino acids 93 to 212 of preproendothelin-1.
2. The method as claimed in claim 1, wherein the determination in biological fluid is effected with the aid of an immunoassay which operates with at least one marked antibody which specifically recognizes only the peptide fragment to be determined.
3. The method as claimed in claim 2, wherein the immunoassay is a competitive immunoassay or a sandwich immunoassay.
4. The method as claimed in claim 1, wherein those C-terminal fragments of preproendothelin-1 are determined which are recognized by antibodies which bind to peptides which correspond to peptide sequences in the range of the amino acids 168 to 212 (SEQ ID NO:7) of preproendothelin-1.

5. The method as claimed in claim 4, wherein pairs of antibodies which bind to two different peptide sequences which are selected from peptide sequences having the amino acids 168-181, 184-203 and 200-212 of preproendothelin-1 are used for determining a C-terminal fragment having amino acids 168 to 212 of preproendothelin-1 (SEQ ID NO:7).
6. The method as claimed in any of the preceding claims, which is a method for the quantitative or for the semiquantitative determination of the peptide fragments to be determined.
7. The method as claimed in claim 6, which is an immunochromatographic point-of-care test or another accelerated test.
8. The method as claimed in any of claims 4 to 7, wherein the antibodies used for the determination are monoclonal and/or affinity-purified polyclonal antibodies.
9. The method as claimed in any of claims 4 to 8, wherein antibodies which are obtained by immunizing an animal with an antigen which contains a synthetic peptide which is selected from the peptides (SEQ ID NO:4), (SEQ ID NO:5) and (SEQ ID NO:6) are used.
10. The method as claimed in any of claims 4 to 9, wherein two antibodies are used for the determination, one of which is marked and the

other is bound to a solid phase or can be selectively bound to a solid phase.

- 5 11. The method as claimed in any of claims 4 to 9,
determination, both of which are present in
dispersed form in the liquid reaction mixture, a
first marking component which is part of a marking
10 distinction or amplification being bound to the
first antibody, and the second marking component
of this marking system being bound to the second
antibody so that, after binding of both antibodies
to the peptide fragment to be detected, a
15 measurable signal which permits detection of the
resulting sandwich complexes in the measuring
solution is generated.
- 20 12. The method as claimed in claim 11, wherein the
marking system comprises rare earth cryptates or
chelates in combination with a fluorescent or
chemiluminescent dye, in particular of the cyanine
type.
- 25 13. The method as claimed in any of claims 1 to 12,
which is used for diagnosis, for determination of
severity and for prognosis and for monitoring the
therapy in the course of sepsis.
- 30 14. The method as claimed in claim 13, which is
carried out as part of a multiparameter
determination, in which at least one further
parameter relevant to sepsis diagnosis is

determined simultaneously.

15. The method as claimed in claim 14, wherein the further parameter or parameters relevant for sepsis diagnosis is or are selected from the group which consists of anti-ganglioside antibodies, the proteins calcitonin, CA 125, CA 19-9, S100B, S100A proteins, LASP-1, soluble cytokeratin fragments, in particular CYFRA 21, TPS and/or soluble cytokeratin-1 fragments (sCY1F), the peptides inflammin and CHP, fragments of the prohormones pro-ANP, pro-BNP or pro-ADM, glycine-N-acyltransferase (GNAT), carbamoylphosphate synthetase 1 (CPS 1) and C-reactive protein (CRP) or fragments thereof.
16. The method as claimed in any of claims 1 to 12, which is used in the area of cardiac diagnostics.
17. The method as claimed in claim 16, which is carried out as part of a multiparameter determination, in which further parameters relevant to cardiac diagnostics are determined simultaneously.
18. The method as claimed in any of claims 1 to 12, which is used in the area of cancer diagnostics.
19. The method as claimed in claim 18, which is carried out as part of a multiparameter determination, in which further parameters relevant to cancer diagnostics are determined simultaneously.

20. An antibody which binds specifically to peptides which consist of the amino acid sequences which correspond to the amino acids 168-181, 184-203 and 200-212 of preproendothelin-1.

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21. The antibody as claimed in claim 20, which is an affinity-purified polyclonal antibody or monoclonal antibody.

10 22. A kit for carrying out a method as claimed in any of claims 1 to 19, which comprises at least: (a) a first antibody as claimed in either of claims 20 and 21, (b) a second, different antibody as claimed in either of claims 20 and 21, one of the
15 antibodies being marked and the other being immobilized or immobilizable, and (c) a standard peptide which has an amino acid sequence which comprises at least the amino acids 168-203 or 168-212 of preproendothelin.

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23. The kit as claimed in claim 22, wherein the immobilized antibody is present in immobilized form on the walls of a test tube (CT).